MECHANISMS OF PIERCE'S DISEASE TRANSMISSION IN GRAPEVINES: XYLEM PATHWAYS AND MOVEMENT OF XYLELLA FASTIDIOSA

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ABSTRACT

This progress report shows that open pathways likely exist for *Xylella fastidiosa* (*Xf*) movement across grafts in grape stems via xylem pathways. Studies thus far have been conducted on grafted and non-grafted three-year-old *Vitis vinifera* cv. Chardonnay plants. The movement of air was used to determine if open xylem conduits were present through grafts into canes, and the length of these pathways was measured. It was determined that connections via xylem vessels are generally about twice as long in non-grafted plants (450 mm) compared with grafted plants (225 mm). Current investigations are underway with dilute use latex paint and tagged *Xf* to understand the pathways for bacterial movement across grafts.

INTRODUCTION

Grapes are one of the important crop plants in which the shoots of one variety are grafted to root stocks of another to generate plants with the desired characteristics of both. Reports have clearly shown the presence of long, open xylem conduits that connect stems to leaves in chardonnay (Thorne et al., 2006; Chatelet et al, in press). Anatomical studies have also indicated that *Xylella fastidiosa* (*Xf*) appears to be primarily restricted to xylem vessels in canes, however little is known about the vessels, and subsequently the movement of *Xf* across grafts. The capacity for *Xf* to move in plants differs among species ranging from generally unrestricted throughout the major organs, to only a few centimeters from the original inoculation point. The objectives of this study are to examine the connection of vessels from canes into stems through grafts, and determine if it is possible for *Xf* movement to occur freely across these grafts. To meet these objectives a strategy of air and latex paint are being implemented to study open anatomical systems, and most importantly the use of *Xf* to examine movement across grafts.

OBJECTIVES

- 1. Conduct a study of connections in grafted *Vitis vinifera* cv. Chardonnay, and determine if open vessel systems allow movement of *Xf* across grafts via air pressure.
- 2. Conduct an anatomical study of connections in grafted *Vitis vinifera* cv. Chardonnay, and determine if open vessel systems allow movement of *Xf* across grafts with latex paint.
- 3. Use PCR to determine the presence of Xf across graft unions after inoculation at known positions relative to the graft.

RESULTS

Following inoculation in grapevine, Xf moves in the nutrient poor xylem vessels and eventually causes disease symptoms that result plant death by unknown mechanisms. Previously, reports from our labs have indicated that bacteria can move freely in canes and from petioles into leaves during a systemic infection process. It is of interest to determine the movement across grafts to clarify movement into stems and possibly into root systems. Our preliminary results indicate that the graft unions of $Vitis\ vinifera\ cv$. Chardonnay do indeed contain continuous vessels; however, the open system length into canes is about $\frac{1}{2}$ of that when compared with non-grafted plants of the same cultivar (Figure 1A). Measurements collected of cane length and associated open conduits appeared significantly different between grafted and non-grafted plants (Figure 1B). Differences were not found to be significant in stem length between grafted and non-grafted plants (Figure 1C). These results indicate that graft unions would not be an impediment to bacterial movement, and that Xf would be able to move further distances across in non-grafted areas of the plant because of the presence of continuous vessels. This study of the xylem structure will be further evaluated with current studies to determine the connective pathway of air movement by latex paint, and confirm that Xf can be moved through the vessels in the presumed transpiration stream.

CONCLUSIONS

From our preliminary results, graft unions in stems do not appear to restrict the movement of Xf in $Vitis\ vinifera\ cv$. Chardonnay. Although the length of open vessels is reduced by about $\frac{1}{2}$, open vessels cross the graft union as determined by air movement. However, the numbers of vessels that cross the graft are less than 10% in distribution when compared with non-grafted plants (data not shown). In order for Xf to move from a cane or leaf across a graft it would need to be inoculated into a vessel that happens to extend through the graft union, or the bacteria would need to degrade membranes to move into adjacent vessels through bordered pits. Current studies with paint and PCR detection of Xf will confirm these results.

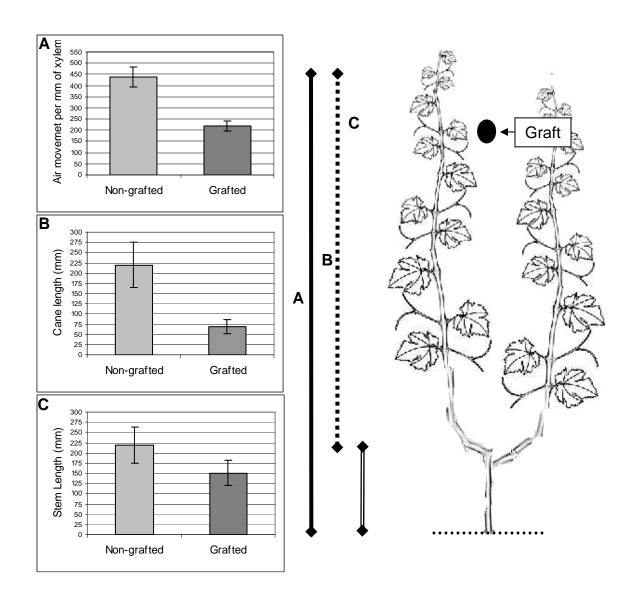


Figure 1. Air movement through graft system. Results indicate that the graft unions of *Vitis vinifera* cv. Chardonnay contain continuous vessels. (A) The open system length into canes is about ½ of that when compared with non-grafted plants of the same cultivar. (B, C) Cane length measurements are significantly different between grafted and non-grafted plants, however differences were not found to be significant in stem length between grafted and non-grafted plants (C).

REFERENCES

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MECHANISMS OF PIERCE'S DISEASE TRANSMISSION IN GRAPEVINES: THE XYLEM PATHWAYS AND MOVEMENT OF XYLELLA FASTIDIOSA. XYLEM STRUCTURE AND CONNECTIVITY IN GRAPEVINE SHOOTS PROVIDES A PASSIVE MECHANISM FOR THE SPREAD OF BACTERIA IN GRAPE PLANTS INFECTED WITH PIERCE'S DISEASE

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ABSTRACT

This progress report shows that there is a difference in the structure of xylem pathways available for *Xylella fastidiosa* (*Xf*) movement in host plants known to support systemic bacterial movement and those that don't. In addition to a reduced interorgan connectivity, non-systemic species also show a shorter path available for bacterial movement in the leaves. However, systemic and non-systemic species show similar rates of tylose formation, signifying that tyloses don't seem to be responsible for the lack of *Xf* movement in the non-systemic plants. To be more conclusive, more xylem characteristics from the different hosts are being examined

INTRODUCTION

Xylella fastidiosa (*Xf*) capacity to move in plants differs greatly among species (Purcell, 2004), ranging from moving everywhere in the stem and leaves to only a few centimeters from the infection point. Our lab showed the presence of long xylem conduits from stem to leaves in grape cultivars chardonnay and cowart (Thorne et al., 2006; Chatelet et al, in press) and we recently reported that these conduits seemed to be shorter in alternate hosts in which bacterial movement is limited. A higher number of tracheids, shorter and narrower vessels, spatial organization of the vessel and of the paratracheal parenchyma cells could be a passive strategy to limit bacterial movement. Another strategy of the non-systemic species could be to confine the bacteria to a limited area by a more timely production of tyloses or, in the case of asymptomatic species showing systemic bacterial movement, to limit the population size under a harmless threshold. The objectives of this study are to carefully study the comparative anatomy of different species of plants which support a range of *Xf* population sizes and movement characteristics. Our hope is to understand how the xylem network might control bacterial movement in susceptible plants.

OBJECTIVES

- 1. Conduct an anatomical comparison of plant species that support high, medium and low population sizes of Xf.
- 2. Conduct an anatomical comparison of plant species that show systemic movement of Xf vs. those that do not.

RESULTS

A range of species was examined: with a high infection rate, high bacterial population and showing systemic movement: *Vitis vinifera* cv. Chardonnay and *Vitis vinifera* cv. Cabernet sauvignon; one species with a high infection rate, medium bacterial population and showing systemic movement: *Ipomoea purpurea* (morning glory), *Vinca major* (periwinkle), *Citrus sinensis* (Orange), *Prunus anygdalus* (Almond), and species showing non-systemic movement: *Alnus rhombifolia* (white alder), *Umbellularia californica* (california laurel), *Artemisia douglasiana* (mugwort) and *Chenopodium quinoa* (quinoa), *Datura wrightii* (datura), *Eucalyptus globules* (eucalyptus).

Stem-petiole-leaf lamina connectivity - Grape shoots have open xylem conduits that allow the passive movement of GFP-Xf from the stem to 50-60% of the leaf length through the primary xylem (Rost et al., PD symposium report 2005; Chatelet et al., in press). The xylem of several different plant species harboring Xf was examined using air and paint injection to determine if similar xylem conduits exist. When loaded at the base of the petiole, air and paint traveled to various extents into the leaf blade of all examined species (Figure 1).